201-14714

Crompton

Product Safety and Regulatory Affairs

August 27, 2003

Marianne L. Horinko
Acting Administrator
U.S. Environmental Protection Agency
P.O. Box 1473
Merrifield, VA 22116

Attn: Chemical Right-To-Know Program

Dear Administrator Horinko,

Crompton Corporation is submitting the enclosed Robust Summary and Test plan for the following chemical:

4-(1-methyl-1-phenylethyl)-N-[4-(1-methyl-1-phenylethyl)phenyl]aniline (CAS # 10081-67-1)

If you have any questions, please contact me at 203-573-3390 or e-mail to mark_thomson@cromptoncorp.com

Sincerely

Dr. Mark A. Thomson
Manager, Toxicology & International Product Registration
Crompton Corporation
Middlebury, CT 06749
USA

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A Public Commitment

201-14714A

HIGH PRODUCTION VOLUME (HPV) CHEMICAL CHALLENGE PROGRAM

TEST PLAN

For

4,4'-Bis(alpha, alpha-dimethylbenzyl)diphenylamine

CAS No. 10081-67-1

Submitted to the US EPA

BY

Crompton Corporation.

Table of Contents

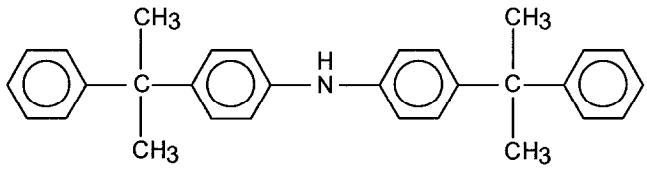
Test Plan for 4,4'-bis(alpha,alpha-dimethylbenzyl)diphenylamine

1. General Information

1.1 CAS Number: 10081-67-1

1.2 Molecular Weight: 405.59

1.3 Structure and formula: $C_{30}H_{31}N$



1.4 Introduction

4,4'-Bis(alpha, alpha-dimethylbenzyl)diphenylamine is an antioxidant, used as a thermal stabiliser in the plastics industry (including specific uses in the production of polyolefins, styrenics, polyols, hot melt adhesives, lubricants and polyamines).

2. Review of Existing Data and Development of Test Plan

Crompton Corporation has undertaken a comprehensive evaluation of all relevant data on the SIDS endpoints of concern for 4,4'-bis(alpha,alpha-dimethylbenzyl)diphenylamine.

The availability of the data on the specific SIDS endpoints is summarized in Table 1. Table 1 also shows data gaps that will be filled by additional testing.

Table 1: Available adequate data and proposed testing on 4,4'-bis(alpha,alpha-dimethylbenzyl)diphenylamine

CAS No. 10081-67-1	Information Available?	GLP	OECD Study?	Other Study?	Estimation Method?	Acceptable?	SIDS Testing required?
	Y/N	Y/N	Y/N	Y/N	Y/N	Y/N	Y/N
Physicochemical							
Melting Point	Y						N
Boiling Point	Y				Y	Y	N
Vapour Pressure	Y	N		Y		Y	N
Water Solubility	Y	N		Y		Y	N
Partition Coefficient (Kow)	Y				Y	Y	N
Environmental Fate							
Biodegradation	Y				Y	Y	N
Hydrolysis	N						N
Photodegradation	Y				Y	Y	N
Transport and Distribution between Environmental Compartments	Y				Y	Y	N
Ecotoxicology							
Acute Fish	Y				Y	Y	N
Acute Daphnia	Y				Y	Y	N
Acute Algae	Y				Y	Y	N
Toxicology							
Acute Oral	Y			Y		Y	N
Repeat Dose toxicity	N						Y
Genetic toxicity – Gene mutation	Y			Y		Y	N
Genetic toxicity – Chromosome aberration	N						Y
Reproductive toxicity	N						Y
Developmental toxicity/teratogenicity	N						Y

A. Evaluation of Existing Physicochemical Data and Proposed Testing

1. Melting Point

The Safety Data Sheet for 4,4'-bis(alpha, alpha-dimethylbenzyl)diphenylamine quotes a melting point of 95°C.

2. Boiling Point

The Safety Data Sheet for 4,4'-bis(alpha, alpha-dimethylbenzyl)diphenylamine reports an autoflammability temperature of 298°C. This suggests that the substance will not reach the boiling temperature, calculated to be 507.8 °C using MPBPWIN v1.40.

3. Vapour Pressure

The vapour pressure of 4,4'-bis(alpha, alpha-dimethylbenzyl)diphenylamine was measured to be 6.67 hPa at 20°C using a method similar to OECD Method 104.

4. Water Solubility

The water solubility of 4,4'-bis(alpha, alpha-dimethylbenzyl)diphenylamine was measured to be 7 mg/L.

5. Partition Coefficient

The partition coefficient (i.e. Kow) for 4,4'-bis(alpha, alpha-dimethylbenzyl)diphenylamine was calculated as log Kow = 8.51 using KOWWIN v1.66.

Summary of Physicochemical Properties Testing: Existing data for melting point, boiling point, vapour pressure, partition coefficient and water solubility are considered to fill these endpoints adequately.

B. Evaluation of Existing Environmental Fate Data and Proposed Testing

1. Biodegradation

The biodegradability of the chemical has been estimated using Biowin v4.00 and the results indicate the chemical not to be readily biodegradable.

2. Hydrolysis

There are no hydrolysable groups in the chemical structure, and the substance is predicted to be hydrolytically stable. In addition, hydrolysis testing of poorly soluble substances (i.e. solubility <10 mg/l) is technically difficult. Therefore, no testing will be performed to fulfill this endpoint.

3. Photodegradation

The potential for photodegradation of 4,4'-bis(alpha, alpha-dimethylbenzyl) diphenylamine has been estimated using the AOPWIN v1.90, and indicated atmospheric oxidation via OH radicals reaction with a half-life of 0.64 hours.

4. Transport and Distribution between Environmental Compartments

An Epiwin Level III Fugacity Model calculation has been conducted for 4,4'-bis(alpha, alpha-dimethylbenzyl)diphenylamine and indicates distribution mainly to sediment for emissions of 1000 kg/hr simultaneously to air water and soil compartments.

Summary of Environmental Fate Testing: Existing data for photodegradation and transport and distribution between environmental compartments are considered to fill these endpoints

adequately. The chemical contains no hydrolysable or biodegradable groups, therefore no hydrolysis or biodegradation testing is proposed.

C. Evaluation of Existing Ecotoxicity Data and Proposed Testing

1. Acute Toxicity to Fish

   Estimation using ECOSAR v0.99g gives an LC_{50} (96 h) of 0.00023 mg/L.

2. Acute Toxicity to Algae

   Estimation using ECOSAR v0.99g gives an LC_{50} (96 h) of 0.000349 mg/L.

3. Acute Toxicity to Daphnia

   Estimation using ECOSAR v0.99g gives an LC_{50} (48 h) of 0.00038 mg/L.

Summary of Ecotoxicity Testing: The chemical belongs to the Ecosar class of neutral organics. The predicted values for acute toxicity to fish, daphnia and algae are regarded as being valid for this material and no testing is proposed.

D. Evaluation of Existing Human Health Effects Data and Proposed Testing

1. Acute Oral Toxicity

   The acute oral toxicity has been determined (similar to OECD 401, rat, non-GLP), with a reported LD50 value of > 10,000 mg/kg b.w. Data from a closely related analogue of the sponsored chemical (styrenated N-phenyl-benzenamine, CAS # 68442-68-2) indicates an oral LD50 >500 - >20,000 mg/kg. It is proposed that read across from this analogue is valid and reduces concern over the acute toxicity of 4,4'-bis(alpha,alpha-dimethylbenzyl)diphenylamine, therefore no further testing will be performed.

2. Skin Irritation

   This non-SIDS endpoint has been evaluated (similar to OECD 404, rabbit, non-GLP). The substance was not classified as irritating to skin.

3. Eye Irritation

   This non-SIDS endpoint has been evaluated (rabbit, non-GLP). The substance was not classified as an eye irritant.

4. Repeat Dose Toxicity

   The repeat dose toxicity of 4,4'-bis(alpha,alpha-dimethylbenzyl)diphenylamine will be determined using OECD Method 422.

5. Genotoxicity

4,4'-Bis(alpha,alpha-dimethylbenzyl)diphenylamine was determined to be non-mutagenic in an Ames reverse mutation assay (Ames test, S. typhimurium strains TA98, TA100, TA1535, TA1537, TA1538, GLP).

The *in vitro* cytogenicity of 4,4'-bis(alpha,alpha-dimethylbenzyl)diphenylamine will be determined using OECD Method 473.

6. Reproductive and Developmental Toxicity

The developmental and reproductive toxicity of 4,4'-bis(alpha,alpha-dimethylbenzyl)diphenylamine in rat will be determined using OECD Method 422.

Summary of Human Health Effects Testing: The repeat dose toxicity combined with the developmental and reproductive toxicity will be evaluated using OECD Method 422. The potential to cause in vitro chromosomal aberrations will be determined using OECD Method 473. The existing data for acute oral toxicity and mutagenicity in a bacterial system are considered to fulfil these endpoints adequately and no further testing will be undertaken. 4,4'-Bis(alpha, alpha-dimethylbenzyl)diphenylamine has been shown to be neither a skin nor an eye irritant.

3. Evaluation of Data for Quality and Acceptability

The collected data were reviewed for quality and acceptability following the general US EPA guidance [2] and the systematic approach described by Klimisch et al [3]. These methods include consideration of the reliability, relevance and adequacy of the data in evaluating their usefulness for hazard assessment purposes. This scoring system was only applied to ecotoxicology and human health endpoint studies per EPA recommendation [4]. The codification described by Klimisch specifies four categories of reliability for describing data adequacy. These are:

(1) Reliable without restriction: Includes studies or data complying with Good Laboratory Practice (GLP) procedures, or with valid and/or internationally accepted testing guidelines, or in which the test parameters are documented and comparable to these guidelines.

(2) Reliable with Restrictions: Includes studies or data in which test parameters are documented but vary slightly from testing guidelines.

(3) Not Reliable: Includes studies or data in which there are interferences, or that use non-relevant organisms or exposure routes, or which were carried out using unacceptable methods, or where documentation is insufficient.

(4) Not Assignable: Includes studies or data in which insufficient detail is reported to assign a rating, e.g. listed in abstracts or secondary literature.

4. References

[1] US EPA, EPI Suite Software, 2000

[2] USEPA (1998). Guidance for Meeting the SIDS Requirements (The SIDS Guide). Guidance for the HPV Challenge Program. Dated 11/2/98.

[3] Klimisch, H.-J., et al (1997). A Systematic Approach for Evaluating the Quality of Experimental Toxicological and Ecotoxicological Data. Regul. Toxicol. Pharmacol. 25:1-5

[4] USEPA (1999). Determining the Adequacy of Existing Data. Guidance for the HPV Challenge Program. Draft dated 2/10/99.

201-14714B

I U C L I D

Data Set

Robust Summaries

Existing Chemical	:	ID: 10081-67-1
Memo	:	Crompton Corporation US HPV
CAS No.	:	10081-67-1
EINECS Name	:	4-(1-methyl-1-phenylethyl)-N-[4-(1-methyl-1-phenylethyl)phenyl]aniline
EC No.	:	233-215-5
Molecular Formula	:	C30H31N

Printing date	:	25.06.2003
Revision date	:	
Date of last update	:	25.06.2003
Number of pages	:	13

2.1 MELTING POINT

Value	:	95 °C
Sublimation	:	
Method	:	other: no data
Year	:	2001
GLP	:	
Test substance	:	Chemical name: 4,4'-Di-(alpha, alpha-dimethylbenzyl) diphenylamine (CAS No. 10081-67-1) Trade name: Naugard 445 Source: Crompton Corporation
Reliability	:	(2) valid with restrictions

28.03.2003 (2)

2.2 BOILING POINT

Value	:	= 507.1 °C at
Decomposition	:	
Method	:	other: estimated using MPBPWIN v 1.40
Year	:	2002
GLP	:	
Test substance	:	Chemical name: 4,4'-Di-(alpha, alpha-dimethylbenzyl) diphenylamine (CAS No. 10081-67-1)
Remark	:	The safety data sheet for the substance reports an autoflammability temperature of 298°C, hence it will decompose before reaching its estimated boiling point.
Reliability	:	(2) valid with restrictions

23.12.2002 (2) (7)

2.4 VAPOUR PRESSURE

Value	:	= 6.67 hPa at 20 °C
Decomposition	:	
Method	:	other (measured): similar to OECD 104
Year	:	1989
GLP	:	no
Test substance	:	Trade name: Naugard 445 Chemical name: 4,4'-Di-(alpha, alpha-dimethylbenzyl) diphenylamine (CAS No. 10081-67-1) Source: Uniroyal Chemical Company, Inc. Purity: >99% Lot No.: C-9-E29203
Remark	:	Measurements were made using the standard flask and manometer vapor pressure apparatus. After evacuating all vapor at -30°C, the sample was equilibrated at the various temperatures in a constant temperature bath.
Reliability	:	(2) valid with restrictions

28.03.2003 (5)

2.5 PARTITION COEFFICIENT

Partition coefficient	:	octanol-water

Log pow	:	= 8.51 at °C
pH value	:	
Method	:	other (calculated): KOWWIN v1.66
Year	:	2002
GLP	:	no
Test substance	:	Chemical name: 4,4'-Di-(alpha, alpha-dimethylbenzyl) diphenylamine (CAS No. 10081-67-1)
Reliability	:	(2) valid with restrictions

28.03.2003 (7)

2.6.1 SOLUBILITY IN DIFFERENT MEDIA

Solubility in	:	Water
Value	:	= 7 mg/l at °C
pH value	:	
concentration	:	at °C
Temperature effects	:	
Examine different pol.	:	
pKa	:	at 25 °C
Description	:	
Stable	:	
Deg. product	:	
Method	:	other
Year	:	1986
GLP	:	no data
Test substance	:	Trade name: Naugard 455 Chemical name: 4,4'-Di-(alpha, alpha-dimethylbenzyl) diphenylamine (CAS No. 10081-67-1) Source: Uniroyal Chemical Company, Inc. Purity: 98% Lot No.: 6C302
Method	:	Excess substance added to 100 mL of water (HPLC grade) and sonicated for 4 hours, followed by standing for 1 hour to allow the solution to return to room temperature. The solution was filtered through Whatman #5 filter paper and diluted 1:1 with methanol prior to analysis using GC.
Result	:	Solvent: Solubility: Water 7 mg/l 3% Acetic acid/water 6 mg/l 8% Ethanol/water 8.5 mg/l Heptane 7400 mg/l
Reliability	:	(2) valid with restrictions

23.12.2002 (4)

3.1.1 PHOTODEGRADATION

Type	:	air
Light source	:	
Light spectrum	:	nm
Relative intensity	:	based on intensity of sunlight
DIRECT PHOTOLYSIS		
Halflife t1/2	:	.6 hour(s)
Degradation	:	% after
Quantum yield	:	
Deg. product	:	
Method	:	other (calculated): Estimation using AOPWIN v1.90
Year	:	
GLP	:	
Test substance	:	Chemical name: 4,4'-Di-(alpha, alpha-dimethylbenzyl) diphenylamine (CAS No. 10081-67-1)
Test condition	:	12 hr day, 1.5E6 OH/cm3
Reliability	:	(2) valid with restrictions

28.03.2003 (7)

3.3.1 TRANSPORT BETWEEN ENVIRONMENTAL COMPARTMENTS

Type	:	fugacity model level III
Media	:	
Air	:	% (Fugacity Model Level I)
Water	:	% (Fugacity Model Level I)
Soil	:	% (Fugacity Model Level I)
Biota	:	% (Fugacity Model Level II/III)
Soil	:	% (Fugacity Model Level II/III)
Method	:	other: EPIWIN Level III Fugacity Model
Year	:	2003
Test substance	:	Chemical name: 4,4'-Di-(alpha, alpha-dimethylbenzyl) diphenylamine (CAS No. 10081-67-1)
Test condition	:	Henry's Law Constant: 0.382 atm-m3/mole (Henrywin program) Vapor pressure: 5.01 mmHg (experimental) Log Kow: 8.51 (Kowwin program) Soil Koc: 1.33E+8 (calc by model) Melting point: 95 °C (experimental) 1000 kg/hr emissions to air, water and soil compartments.

	Mass Amount (percent)	Half-life (hr)	Emissions (kg/hr)
Air	0.0244	1.28	1000
Water	2.43	1.44E+3	1000
Soil	27.8	1.44E+3	1000
Sediment	69.7	5.76E+3	0

	Fugacity (atm)	Reaction (kg/hr)	Advection (kg/hr)	Reaction (percent)	Advection (percent)
Air	1.1E-12	985	18.2	32.8	0.606
Water	3.95E-9	87.2	181	2.91	6.04
Soil	3.41E-11	999	0	33.3	0
Sediment	3.85E-9	626	104	20.9	3.47

Persistence time: 2.49E+3 hr
Reaction time: 2.77E+3 hr
Advection time: 2.46E+4 hr
Percent reacted: 89.9
Percent advected: 10.1

Half-lives (hr), (based upon Biowin (ultimate) and Aopwin):
Air: 1.279
Water: 1440
Soil: 1440
Sediment: 5760
Biowin estimate: 1.788 (months)

Advection times (hr):
Air: 100
Water: 1000
Sediment: 5E+4

Reliability : (2) valid with restrictions
28.03.2003 (7)

3.5 BIODEGRADATION

Type	:	aerobic
Inoculum	:	
Deg. product	:	
Method	:	other: Estimation using BIOWIN v4.00
Year	:	2003
GLP	:	
Test substance	:	Chemical name: 4,4'-Di-(alpha, alpha-dimethylbenzyl) diphenylamine (CAS No. 10081-67-1)
Result	:	MITI Linear Biodegradation Probability: -0.367 MITI Non-linear Biodegradation Probability: 0.0006 The chemical is predicted to be not readily biodegradable.
Reliability	:	(2) valid with restrictions

28.03.2003 (7)

4.1 ACUTE/PROLONGED TOXICITY TO FISH

Type	:	
Species	:	
Exposure period	:	96 hour(s)
Unit	:	mg/l
LC50	:	.00023
Method	:	other: Estimation using Ecosar v0.99g
Year	:	2003
GLP	:	
Test substance	:	Chemical name: 4,4'-Di-(alpha, alpha-dimethylbenzyl) diphenylamine (CAS No. 10081-67-1)
Test condition	:	Log Kow: 8.51 (KOWWIN estimate) MPt: 95°C (measured) Water solubility: 7 mg/L (measured)
Reliability	:	(2) valid with restrictions

28.03.2003 (7)

4.2 ACUTE TOXICITY TO AQUATIC INVERTEBRATES

Type	:	
Species	:	Daphnia sp. (Crustacea)
Exposure period	:	48 hour(s)
Unit	:	
EC50	:	.00038
Method	:	other: Estimation using Ecosar v0.99g
Year	:	2003
GLP	:	
Test substance	:	Chemical name: 4,4'-Di-(alpha, alpha-dimethylbenzyl) diphenylamine (CAS No. 10081-67-1)
Test condition	:	Log Kow: 8.51 (KOWWIN estimate) MPt: 95°C (measured) Water solubility: 7 mg/L (measured)
Reliability	:	(2) valid with restrictions

28.03.2003 (7)

4.3 TOXICITY TO AQUATIC PLANTS E.G. ALGAE

Species	:	
Endpoint	:	
Exposure period	:	96 hour(s)
Unit	:	mg/l
EC50	:	.000349
Method	:	other: Estimation using Ecosar v0.99g
Year	:	2003
GLP	:	
Test substance	:	Chemical name: 4,4'-Di-(alpha, alpha-dimethylbenzyl) diphenylamine (CAS No. 10081-67-1)
Test condition	:	Log Kow: 8.51 (KOWWIN estimate) MPt: 95°C (measured) Water solubility: 7 mg/L (measured)
Reliability	:	(2) valid with restrictions

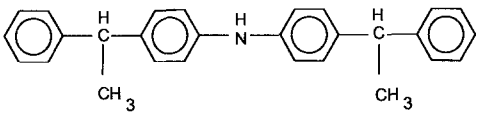
28.03.2003 (7)

5.1.1 ACUTE ORAL TOXICITY

Type	:	LD50
Value	:	> 10000 mg/kg bw
Species	:	rat
Strain	:	other: Holtzman
Sex	:	male
Number of animals	:	30
Vehicle	:	other: Corn oil
Doses	:	215, 464, 1000, 2150, 4640 and 10000 mg/kg bw
Method	:	other: similar to OECD 401
Year	:	1964
GLP	:	no
Test substance	:	Trade name: EPRA Chemical name: 4,4'-Di-(alpha, alpha-dimethylbenzyl) diphenylamine Source: Naugatuck Chemical
Method	:	Dose: Single by gavage Post dose observatation period: 14 days Weight range at start of study: 216-300 g
Result	:	No mortalities occurred at any dosage level tested. The animals at each dosage level generally exhibited normal behaviour and appearance during the 14-d observation period. Average body weight gains for the rats at each dosage level were within normal limits for rats of the age, sex and strain used in this study. Gross autopsies performed on the rats at termination showed no significant gross pathology.
Conclusion	:	The acute oral LD50 of EPRA for male albino rats is >10000 mg/kg bw.
Reliability	:	(2) valid with restrictions No details of sample purity or Lot No.

06.01.2003 (3)

Type	:	LD50
Value	:	> 20000 mg/kg bw
Species	:	rat
Strain	:	
Sex	:	
Number of animals	:	25
Vehicle	:	other: corn oil
Doses	:	2500, 5000, 10000, 20000, 40000 mg/kg
Method	:	other
Year	:	1976
GLP	:	no
Test substance	:	Chemical name: Benzenamine, N-phenyl-, styrenated CAS #: 68442-68-2

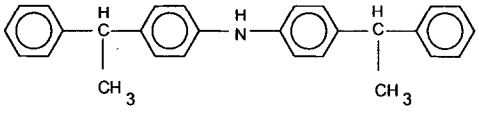


Method	:	Dose: Single oral in 25% corn oil solution Post dose observatation period: 14 days

The test parameters were based on a known and well established procedure for the time period.

Result : Two of the five animals died at the dosages of 20000 and 40000 mg/kg.
Conclusion : The LD50 of this analogue of the sponsored chemical was > 20000 mg/kg b.w.
Reliability : (2) valid with restrictions
06.01.2003 (1)

Type : LD50
Value : > 500 mg/kg bw
Species : rat
Strain : Sprague-Dawley
Sex : male/female
Number of animals : 10
Vehicle : other: corn oil
Doses : 500 mg/kg
Method : other: US Department of Transportation Regulations, 49CFR173.132 (1992)
Year : 1993
GLP : yes
Test substance : Chemical name: Benzenamine, N-phenyl-, styrenated
CAS #: 68442-68-2



Method : Dose: Single by gavage
Post dose observatation period: 14 days
Result : No animals died during the 14 day observation period. No significant clinical findings and no significant impairment on body weight gains were noted in either the male or female rats. No abnormal tissues were note in any animals upon necropsy.
Conclusion : The LD50 of this analogue of the sponsored chemical was > 500 mg/kg b.w.
Reliability : (2) valid with restrictions
06.01.2003 (1)

5.2.1 SKIN IRRITATION

Species : rabbit
Concentration : .5 g
Exposure : Occlusive
Exposure time : 24 hour(s)
Number of animals : 6
Vehicle : other: Corn oil
PDII :
Result : not irritating
Classification : not irritating
Method : other: similar to OECD 404
Year : 1964
GLP : no
Test substance : Trade name: EPRA
Chemical name: 4,4'-Di-(alpha, alpha-dimethylbenzyl) diphenylamine
Source: Naugatuck Chemical

Result : EPRA produced no gross signs of dermal irritation on intact or abraded skin.

Conclusion	:	Under the conditions of the test, EPRA is not a primary skin irritant.
Reliability	:	(2) valid with restrictions No details of sample purity or Lot No.

23.12.2002 (3)

5.2.2 EYE IRRITATION

Species	:	rabbit
Concentration	:	3 mg
Dose	:	
Exposure time	:	72 hour(s)
Comment	:	not rinsed
Number of animals	:	6
Vehicle	:	none
Result	:	not irritating
Classification	:	not irritating
Method	:	other
Year	:	1964
GLP	:	no data
Test substance	:	Trade name: EPRA Chemical name: 4,4'-Di-(alpha, alpha-dimethylbenzyl) diphenylamine Source: Naugatuck Chemical
Method	:	3 mg of EPRA was applied to the right eye of each of six albino rabbits. The left eye was untreated and served as a control. Each rabbit was examined for eye irritative effects and for gross signs of systemic toxicity at intervals of 24, 48 and 72 hours after application. Irritative effects observed in the eye were scored according to the Draize method.
Result	:	No gross signs of eye irritation were observed at any observation interval following application of EPRA to the eyes of albino rabbits. All rabbits showed normal appearance throughout the study and there was no evidence of systemic toxicity from mucous membrane absorption.
Conclusion	:	EPRA produced no gross signs of eye irritation.
Reliability	:	(2) valid with restrictions No details of sample purity or Lot No.

23.12.2002 (3)

5.4 REPEATED DOSE TOXICITY

5.5 GENETIC TOXICITY 'IN VITRO'

Type	:	Ames test
System of testing	:	Salmonella typhimurium: TA 1535, TA 1537, TA 1538, TA 98 and TA 100
Test concentration	:	+/- S9: 0, 50, 150, 500, 1500 and 5000 µg/plate
Cycotoxic concentr.	:	>5000 µg/plate
Metabolic activation	:	with and without
Result	:	negative
Method	:	other: based on Ames, B.N. et al, Proc. Nat. Acad. Sci. USA (1973), 70, 2281
Year	:	1985

GLP : yes
Test substance : Trade name: Naugard 445
Chemical name: 4,4'-Di-(alpha, alpha-dimethylbenzyl) diphenylamine
Purity: 98 %

Method : Metabolic activation: S9-mix, Rat liver cells, 0.5 ml, Aroclor induced

Statistical Methods: no data

Number of replicates: 3

Positive controls:

N-ethyl-N'-nitro-N-nitrosoguanidine (-S9, TA100 & TA1535)
9-aminoacridine (-S9, TA 1537)
2-nitrofluorene (-S9, TA1538 & TA98)
2-aminoanthracene (+S9, TA98, TA100, TA1535, TA1537 & TA1538)

Negative control: Solvent vehicle

Solvent: Dimethylsulfoxide

Result : The revertant colony counts for Naugard 445 obtained in the dose range finding test are shown in Table 1. Naugard 445 was not toxic towards the tester strains, therefore 5000 µg/plate was chosen as the top dose level in the mutation test.

The mean number of revertant colonies, together with the individual plate counts for Naugard 445 obtained in the mutation test are shown in Table 2. Compound sterility and positive control mutability checks are shown in Table 3.

No substantial increases in revertant colony numbers of any of the five tester strains were observed following treatment with Naugard 445 at any dose level, either in the presence or absence of metabolic activation (S9 mix).

Table 1. Dose range finding test on Naugard 445 - revertant colony counts obtained with *S. typhimurium* strains TA 1535, TA 1537, TA 1538, TA 98 and TA 100

Dose level (ug/plate)	Metabolic activation	Strains of *S. typhimurium*				
		TA 1535	TA 1537	TA 1538	TA 98	TA 100
5000	-	12	10	9	12	120
500	-	4	14	11	15	112
50	-	6	12	4	14	90
5	-	7	12	13	21	90
Solvent	-	16	7	10	12	87
5000	+	9	12	18	23	132
500	+	5	22	11	20	122
50	+	10	19	8	18	99
5	+	8	17	23	14	98
Solvent	+	13	12	15	18	104

Table 2. Naugard 445 - revertant colony counts obtained per plate using *S. typhimurium* strains TA 1535, TA 1537, TA 1538, TA 98 and TA 100

Strain	Dose level (ug/plate)	Without metabolic activation		With metabolic activation	
		Mean revertant colony counts	Individual revertant colony counts	Mean revertant colony counts	Individual revertant colony counts
TA 1535	5000	11	8, 12, 14	11	7, 11, 14
	1500	7	9, 9, 3	10	5, 12, 13
	500	14	16, 16, 9	9	11, 4, 11
	150	12	9, 13, 13	11	9, 9, 15
	50	14	23, 5, 13	9	5, 11, 12
	0	15	15, 15, 16	10	11, 4, 14
	Solvent	14	15, 11, 15	13	10, 15, 13
TA 1537	5000	13	17, 13, 9	19	15, 20, 21
	1500	19	18, 27, 11	14	10, 19, 14
	500	12	20, 9, 6	16	18, 20, 11
	150	16	19, 15, 15	17	15, 19, 18
	50	7	7, 10, 4	18	18, 14, 21
	0	11	12, 12, 10	12	14, 14, 8
	Solvent	8	8, 5, 10	18	21, 19, 14
TA 1538	5000	10	9, 12, 8	22	25, 19, 22
	1500	15	21, 11, 12	14	16, 18, 8
	500	9	13, 3, 12	17	10, 22, 18
	150	11	8, 16, 9	19	23, 17, 18
	50	8	11, 8, 6	16	17, 7, 23
	0	7	7, 4, 10	12	10, 7, 19
	Solvent	9	9, 8, 9	15	20, 12, 13
TA 98	5000	19	18, 18, 21	20	25, 14, 22
	1500	19	17, 20, 20	23	23, 22, 23
	500	15	14, 15, 17	21	28, 16, 20
	150	19	20, 20, 16	23	27, 20, 23
	50	18	14, 21, 19	17	14, 17, 21
	0	22	27, 20, 19	18	15, 21, 19
	Solvent	21	12, 27, 23	23	22, 21, 26
TA 100	5000	101	103, 99, 100	105	104, 97, 115
	1500	87	91, 102, 69	126	121, 121, 136
	500	114	116, 112, 114	130	128, 121, 142
	150	99	109, 83, 106	105	100, 102, 114
	50	105	91, 116, 107	134	127, 128, 146
	0	106	104, 109, 105	119	124, 119, 114
	Solvent	82	107, 67, 72	100	105, 104, 90

Table 3. Mutability and sterility tests with *S. typhimurium* strains TA 1535, TA 1537, TA 1538, TA 98 & TA 100

Strain	Compound	Dose level (ug)	Metabolic activation	Mean revertant colony counts	Individual revertant colony counts
TA 1535	N-ethyl-N'-nitro-N-nitrosoguanidine	5	-	220	199, 232, 229
TA 1537	9-aminoacridine	80	-	2175	2036, 2352, 2138
TA 1538	2-nitrofluorene	2	-	58	51, 76, 47
TA 98		1	-	72	68, 60, 89
TA 100	N-ethyl-N'-nitro-N-nitrosoguanidine	3	-	406	432, 407, 379
TA 1535		2	+	108	114, 105, 106
TA 1537		2	+	54	49, 53, 59
TA 1538	2-aminoanthracene	0.5	+	227	214, 255, 213
TA 98		0.5	+	121	126, 128, 110
TA 100		0.5	+	281	294, 293, 257
-	S-9 mix	500 ul		0	0
-	Naugard 445	5000		0	0

Conclusion : It is concluded that no evidence of mutagenic potential of Naugard 445 was obtained in this bacterial test system at the dose levels used.

Reliability : (1) valid without restriction
Study conducted following recognised test method under GLP.

24.12.2002 (6)

5.8.2 DEVELOPMENTAL TOXICITY/TERATOGENICITY

(1) American Chemistry Council, Rubber and Plastic Additives Panel, HPV Chemical Challenge Program Submission, Substituted Diphenylamines (DPA), December 2001

(2) Crompton Corporation, Naugard 445 Safety Data Sheet, MSDS # C266001, Rev. 6, August 2001

(3) Hill Top Research Institute, Inc., Acute oral, primary skin irritation and eye irritation studies on IVTI, XKIE, FFUU, EPRA and BUTAZATE. 1964.

(4) In-house study conducted by Uniroyal Chemical Company, Inc.'s Chemical Characterization Laboratory, 1986

(5) In-house study conducted by Uniroyal Chemical Company, Inc.'s Chemical Characterization Laboratory, 1989

(6) Jones, E., Fenner, L.A., Thompson, A.L., Huntingdon Research Centre, Ames metabolic activation test to assess the potential mutagenic effect of Naugard 445, HRC Report No. URL 33/851340, 1985

(7) US EPA, EPIWIN v3.10, EPI Suite Software, 2000